

EXPERIMENTAL PAPER

Comparison of chemical composition of the essential oils from different parts of *Thuja occidentalis* L. 'Brabant' and *T. occidentalis* L. 'Smaragd'

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S u m m a r y

Introduction: *Thuja occidentalis* 'Brabant' and *T. occidentalis* 'Smaragd' are widely planted as decoration in parks and as hedges in domestic gardens in Poland. **Objective:** The aim of the study was to determine the yield and the composition of essential oils from different parts of these cultivars. **Methods:** The essential oils were obtained by hydrodistillation, and then analyzed by GC/MS. **Results:** Over seventy compounds were identified. The oils from leaves, twigs with leaves and twigs without leaves of 'Brabant' variety contained mainly α -thujone (61.0–64.4%), β -thujone (10.4–10.7%) and fenchone (7.7–8.0%), whereas in the cone oil there were α -thujone (42.1%), sabinene (11.2%) and β -thujone (10.2%). The main constituents of the oil from leaves and twigs with leaves of 'Smaragd' variety were α -thujone (49.2–49.3%), beyerene (12.8–13.2%) and sabinene (8.9–9.3%). **Conclusions:** The yield and chemical composition of the essential oil of *T. occidentalis* depend on the variety and plant organ. The oil of 'Brabant' variety contained more thujones and less diterpenes in comparison with the oil of 'Smaragd' variety.

Key words: *Thuja occidentalis* 'Brabant', *Thuja occidentalis* 'Smaragd', essential oil composition, α -thujone, β -thujone, beyerene

INTRODUCTION

Thuja occidentalis L. (*Cupressaceae*), commonly known as white cedar or arborvitae, is a coniferous tree with a compact narrowly pyramidal habit, flattened

twigs, evergreen leaves and small cones. The species is native to eastern North America and there is used for timber [1, 2]. In Poland it is widely planted as a decoration in parks and as hedges, screens or to create punctuation mark in domestic gardens. Over 120 cultivar varieties of *T. occidentalis* exist, showing great variation in shape, size and foliage color. The most popular in Poland are *T. occidentalis* 'Brabant' and *T. occidentalis* 'Smaragd' (Emerald) [1-3]:

'Brabant' – 5–8 m height, 120 cm wide, loose columnar habit, fast-growing (50 cm per year), good on the dense hedges but require forming;

'Smaragd' – 3 m height, 60 cm wide, tight pyramidal habit, very thin branches, slower growing (10 cm per year), none-flowering, none-forming cones, good on the dense hedges needing low maintenance (no formation).

T. occidentalis has been used in folk medicine for the treatment of various diseases, such as headache, migraine, cold fever, bronchial catarrh, rheumatism, psoriasis, enuresis, cystitis, amenorrhea, and as contraceptive and abortifacient agent. Ethanolic extract of twigs possesses antibacterial, antifungal, antiviral, antioxidant, anticancer, antirheumatic, antispasmodic, antidiabetic, diuretic, expectorant, hepatoprotective, stimulant, tonic and vermifuge properties. Nowadays, it is applied mainly in homeopathy as mother tincture or dilution. In combination with other plants it is also used in phytotherapy as immunostimulating and antiviral drug in acute and chronic infections of the upper respiratory tract [4-6].

Essential oil of *T. occidentalis* (cedar leaf oil) is commercially produced by hydrodistillation or steam distillation from the foliage in Canada and USA. It is used in pharmaceutical products because of its therapeutic properties, as well as in perfumes, cosmetics, soaps, deodorants, room sprays, detergents, cleansers, disinfectants and insecticides. Due to presence of toxic thujone, which can evoke dizziness and convulsions, dose of the oil must be strictly controlled. Pure thujone is used as active ingredient in the production of nasal decongestants of cough suppressants; because of anticancer activity it may be applied in chemotherapies [7].

Several papers described *T. occidentalis* essential oil, its chemical composition [8-13] and additional biological activities: antimicrobial [14-17] and insecticidal [18-22]. The following factors influence the yield and chemical composition of the oil: origin of the plants [8-12, 14, 18, 19], cultivar variety [13, 15], harvest season [9, 13], technique of oil isolation [8, 9] and time of hydrodistillation [10].

In the present work, the composition of the essential oils from different parts of *T. occidentalis* 'Brabant' and *T. occidentalis* 'Smaragd' is reported. It is the first data on the essential oils composition of these cultivars.

MATERIAL AND METHODS

Plant material

T. occidentalis 'Brabant' and *T. occidentalis* 'Smaragd' were collected in the Łódź Botanical Garden, Poland, in October 2015. The voucher specimens (Tobc

1/2015 and Tocs 1/2015) have been deposited in the Herbarium of the Institute of General Food Chemistry, Lodz University of Technology.

Isolation of the essential oils

Fresh branches were divided into four groups: twigs with leaves, leaves, twigs without leaves and cones. Each sample of fresh plant material (100.0 g) was cut into small pieces (0.5 cm long) and hydrodistilled in a Deryng apparatus for 3 hours to obtain essential oil. Three replicates of each sample were carried out. The oils had pale yellow color and pleasant, intensive herbal-camphoraceous aroma.

Analysis of the essential oils

The chemical composition of the oils were determined by simultaneous GC/FID and GC/MS analyses using a MS-FID splitter (SGE Analytical Science) and a Trace GC Ultra gas chromatograph coupled with a DSQ II mass spectrometer (Thermo Electron Corporation). The apparatus was equipped with an apolar capillary column Rtx-1 (dimethylpolysiloxane), 60 m × 0.25 mm × 0.25 μm film thickness (Restek, Bellefonte, PA, USA). The oven temperature was programmed from 50°C to 300°C, rising 4°C/min. Injector temperature was 280°C, detector temperature 300°C, ion source temperature 200°C, carrier gas helium with constant pressure 300 kPa, ionization voltage 70 eV, mass range 33–420 amu.

Identification and quantification of components

Identification of components was based on comparison of their retention indices RI and their mass spectra MS with those of commercial libraries MassFinder 3.1, NIST 98.2, Wiley Registry of Mass Spectral Data 8th ed. and literature [23].

A quantitative analysis (expressed as percentage of each component) was carried out by peak area normalization measurements without correction factors.

Ethical approval: The conducted research is not related to either human or animal use.

RESULTS AND DISCUSSION

The yields (averages of three replicates) and chemical composition of the essential oils from different parts of *T. occidentalis* 'Smaragd' and *T. occidentalis* 'Brabant' are presented in table 1. The yield of the oil depends on the variety and plant organ. Twigs with leaves of 'Brabant' variety contained 2.5 times more oil

than twigs with leaves of 'Smaragd' variety (1.20% and 0.48%, respectively). In both varieties, leaves contained several times more oil than twigs without leaves (1.21% and 0.12%; 0.50% and 0.09%, respectively). Most of the oil contained cones of 'Brabant' variety (1.50%). According to the literature data, the content of the essential oil in *T. occidentalis* fresh leaves varied between 0.35% [13] to 0.82% [7]. Present study shows that 'Brabant' variety of *T. occidentalis* is much richer in essential oil than previously described plants of this species.

Table 1.

Chemical composition of the essential oils from twigs with leaves (A), leaves (B), twigs without leaves (C), cones (D) of *Thuja occidentalis* L. 'Smaragd' and *T. occidentalis* L. 'Brabant'

Compound	RI	Percentage							
		<i>T. occidentalis</i> 'Smaragd'			<i>T. occidentalis</i> 'Brabant'				
		A	B	C	A	B	C	D	
α -Thujene	925	0.5	0.4	tr	0.2	0.2	0.1	0.5	
α -Pinene	932	0.9	0.9	1.5	0.7	0.8	0.5	4.4	
α -Fenchene	939	0.3	0.2	0.1	0.5	0.4	0.5	0.3	
Camphene	944	0.3	0.3	0.1	0.4	0.4	0.4	0.3	
Sabinene	968	8.9	9.3	3.1	2.8	3.0	0.2	11.2	
β -Pinene	970	0.2	0.2	0.1	0.1	0.1	tr	0.4	
Myrcene	982	1.5	1.5	0.5	0.8	0.8	0.1	1.6	
α -Terpinene	1006	0.4	0.3	0.2	0.2	0.2	0.6	1.0	
<i>p</i> -Cymene	1009	0.2	0.2	0.2	0.3	0.2	1.1	0.3	
Limonene	1020	0.9	0.9	0.6	0.8	0.7	0.8	1.1	
γ -Terpinene	1050	0.7	0.6	0.3	0.4	0.4	0.8	1.6	
<i>trans</i> -Sabinene hydrate	1054	0.4	0.4	0.1	0.3	0.3	0.1	0.6	
Fenchone	1067	5.0	4.9	3.0	7.7	7.7	8.0	7.3	
Terpinolene	1079	0.3	0.3	0.1	0.2	0.2	0.4	0.5	
α -Thujone	1093	49.2	49.3	38.4	61.6	61.0	64.4	42.1	
β -Thujone	1100	3.2	3.6	2.9	10.4	10.7	10.4	10.2	
<i>cis-p</i> -Menth-2-en-1-ol	1108	0.6	0.5	0.3	0.6	0.6	0.3	0.9	
Camphor	1120	0.8	0.8	0.6	0.3	0.4	0.4	0.3	
Neoisothujol	1125	0.2	0.2	0.1	0.2	0.2	0.1	0.4	
Camphene hydrate	1134	0.2	0.2	0.1	0.4	0.2	0.2	0.5	
Thujol	1151	0.4	0.4	0.3	0.6	0.6	0.4	0.3	
Terpinen-4-ol	1163	1.8	1.9	1.6	1.5	1.5	1.2	5.3	
α -Terpineol	1174	0.2	0.2	0.1	0.3	0.3	0.5	0.5	
Bornyl acetate	1269	1.2	1.2	1.7	2.6	2.6	2.7	2.2	
Sabinyl acetate	1276	0.5	0.5	0.7	0.4	0.4	0.5	0.2	
α -Terpinyl acetate	1333	0.7	0.7	1.1	0.8	0.8	0.2	0.8	

Compound	RI	Percentage						
		<i>T. occidentalis</i> 'Smaragd'			<i>T. occidentalis</i> 'Brabant'			
		A	B	C	A	B	C	D
Germacrene D-4-ol	1566	0.5	0.5	0.5	0.1	0.1	0.1	0.2
Caryophyllene oxide	1571	0.4	0.4	4.1	0.1	tr	tr	tr
Humulene epoxide II	1594	0.3	0.2	1.7	tr	tr		tr
<i>epi</i> - α -Muurolool	1626	0.3	0.3	0.8	tr	tr		tr
α -Cadinol	1638	0.3	0.3	0.7	0.1	tr		tr
Rimuene	1896	2.0	2.0	3.1	0.6	0.7	0.2	0.1
Beyerene	1944	13.2	12.8	20.1	1.0	1.1	0.4	0.2
Abietatriene	2040	0.1	0.1	0.6	tr	tr	tr	0.1
Totarol	2249	tr	tr	2.8	0.1	0.1	0.3	0.2
Abietal	2256	tr	tr	1.4				0.1
Oil yield		0.48	0.50	0.09	1.20	1.21	0.12	1.50

RI – retention indices on Rtx-1 column; tr – trace (<0.05%);

compounds with quantity 0.1–0.3% in A-D oils: α -phellandrene, *trans*-*p*-menth-2-en-1-ol, sabina ketone, neothujol, verbenone, *trans*-piperitol, fenchyl acetate, thymol methyl ether, carvacrol methyl ether, *cis*-sabinene hydrate acetate, *trans*-sabinene hydrate acetate, geranyl acetate, (*E*)-cinnamyl acetate, (*E*)- β -caryophyllene, (*E*)-ethyl cinnamate, ethyl (*2E,4Z*)-deca-2,4-dienoate, α -humulene, α -muurolole, γ -cadinene, δ -cadinene, elemol, β -oploponone, 1-*epi*-cubebol, 14-hydroxy- β -caryophyllene, oplopanone, pimara-8(14),15-diene, manoyl oxide, kaur-15-ene, abieta-7,13-diene;

compounds with quantity < 0.1% in A-D oils: tricyclene, 3-carene, cuminyl acetate, γ -muurolole, tridecan-2-one, germacrene D, *epi*-cubebol, α -cadinene, (*E*)-nerolidol

More than seventy compounds representing 97–99% of oils were identified in each, 'Brabant' and 'Smaragd' varieties of *T. occidentalis*. The composition of the oils from leaves, twigs with leaves and twigs without leaves of 'Brabant' variety were similar. The main constituents of the oils were α -thujone (61.0–64.4%), β -thujone (10.4–10.7%) and fenchone (7.7–8.0%). A few components were of concentration 1–3%: sabinene, terpinen-4-ol, bornyl acetate and beyerene, while others amounted lower than 1%. The oil from cones of this variety differed in quantitative composition. It contained mainly α -thujone (42.1%), sabinene (11.2%), β -thujone (10.2%), fenchone (7.3%), terpinen-4-ol (5.3%) and α -pinene (4.4%). This is the first report on the composition of the cone essential oil of *T. occidentalis* species. The oil from leaves and twigs with leaves of 'Smaragd' variety contained α -thujone (49.2–49.3%), beyerene (12.8–13.2%), sabinene (8.9–9.3%), fenchone (4.9–5.0%) and β -thujone (3.2–3.6%) as the main constituents. However, in the oil from twigs without leaves there were α -thujone (38.4%), beyerene (20.1%) and caryophyllene oxide (4.1%).

The chemical profiles of the oils from 'Brabant' and 'Smaragd' varieties of *T. occidentalis* were comparable, having α -thujone as the dominant compound. On the other hand, significant differences between the oils were observed. The oil of 'Brabant' variety contained higher total content of monoterpene ketones α -thujone, β -thujone and fenchone (79.4–82.8%) in comparison with the oil of 'Smaragd' variety (44.3–57.8%), as well as lower content of diterpene hydrocarbons (0.6–1.8%

and 14.9–23.8%, respectively) and monoterpene hydrocarbons (5.5–7.4% and 6.8–15.1%, respectively).

Regarding to the previously reported *T. occidentalis* oils, the composition of 'Brabant' variety oil resembled laboratory distilled leaf oil of *T. occidentalis* (variety not indicated) from Wrocław, Poland, with high level of α -thujone (69.8%), β -thujone (9.5%) and fenchone (7.8%) [19]. In turn, the composition of 'Smaragd' variety oil was similar with the oil of *aurea* variety growing in Gdańsk, Poland, and *malo-nyana* variety from Slovakia, in which lower content of α -thujone (30.4–51.6%) and higher content of diterpenes (11.2–23.8%) were found [13, 15]. Commercial oil of *T. occidentalis* produced in Canada and USA contained mainly α -thujone (45–51%), fenchone (13–15%) and β -thujone (7–9%), but no diterpenes [8, 11, 18]. So, the oil from 'Brabant' variety as well as from 'Smaragd' variety differed in quantitative composition from American oils.

CONCLUSIONS

1. The yield and chemical composition of the essential oil of *T. occidentalis* depend on the variety and plant organ.
2. Twigs of fast-growing *T. occidentalis* 'Brabant' can be a very good source of natural thujones due to high amount of essential oil and high content of thujones in the oil.

Conflict of interest: Authors declare no conflict of interest.

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PORÓWNANIE SKŁADU CHEMICZNEGO OLEJKÓW ETERYCZNYCH Z RÓŻNYCH CZĘŚCI MORFOLOGICZNYCH *THUJA OCCIDENTALIS* L. 'BRABANT' I *T. OCCIDENTALIS* L. 'SMARAGD'

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Streszczenie

Wstęp: *Thuja occidentalis* 'Brabant' i *T. occidentalis* 'Smaragd' są powszechnie uprawiane w Polsce jako rośliny dekoracyjne w parkach i jako żywopłotowe w ogrodach. **Cel:** Celem badań było określenie wydajności i składu chemicznego olejków eterycznych z różnych części morfologicznych tych odmian. **Metodyka:** Olejki eteryczne otrzymano metodą hydrodestylacji, następnie analizowano je metodą GC/MS. **Wyniki:** Zidentyfikowano ponad 70 składników. Olejki z liści, gałązek z liśćmi i gałązek bez liści odmiany 'Brabant' zawierały głównie α -tujon (61,0–64,4%), β -tujon (10,4–10,7%) i fenchon (7,7–8,0%), natomiast olejek z szyszek α -tujon (42,1%), sabinen (11,2%) i β -tujon (10,2%). Głównymi składnikami olejków z liści i gałązek z liśćmi odmiany 'Smaragd' były α -tujon (49,2–49,3%), bejeren (12,8–13,2%) i sabinen (8,9–9,3%). **Wnioski:** Zawartość i skład chemiczny olejków eterycznych zależy od odmiany i części morfologicznych *T. occidentalis*. Olejek z odmiany 'Brabant' zawierał więcej tujonów, a mniej diterpenów w porównaniu z olejkiem z odmiany 'Smaragd'.

Słowa kluczowe: *Thuja occidentalis* 'Brabant', *Thuja occidentalis* 'Smaragd', skład olejku eterycznego, α -tujon, β -tujon, bejeren